

Appendix C



review

Management of Lung Transplant Rejection*

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ALG = antilymphocyte globulin; ATG = antithymocyte globulin; BAL = bronchoalveolar lavage; CMV = cytomegalovirus; FVC = forced vital capacity; HLA = human leukocyte antigen; IL-2 = interleukin-2; MHC = major histocompatibility complex; OB = obliterative bronchiolitis; FFT = pulmonary function test; TBB = transbronchial biopsy

During the past decade lung transplantation has been successfully extended to a variety of advanced lung diseases including pulmonary fibrosis, chronic obstructive pulmonary disease, antitrypsin deficiency emphysema, cystic fibrosis, primary pulmonary hypertension, and selected forms of Eisenmenger's syndrome.^{1,2} In the last few years, activity in isolated lung transplantation has been growing exponentially, while the number of combined heart-lung transplants has been relatively constant (Fig 1). Through August 1992, 1,536 lung transplants had been reported to the St. Louis International Lung Transplant Registry; 1-year and 2-year actuarial survival rates were 68 percent and 60 percent, respectively.² In the United States, 486 lung transplants were performed in 1992, more than twice the number done in 1991,³ and at least 45 centers were active.²

Deaths soon after lung and heart-lung transplantation have been caused primarily by technical and cardiac complications of the operation, but mortality beyond the first month has been related to infection and rejection (Fig 2).¹ The lower respiratory tract is readily exposed to airborne pathogens, while the normal defense mechanisms are breached at many levels in the allografted lung. Mucociliary clearance is diminished, and the cough reflex is depressed because the transplanted lung is denervated. Moreover, the antimicrobial functions of the alveolar macrophage may be impaired.⁴ Hence, the transplanted lung is particularly vulnerable to infection.

After combined heart-lung transplantation, lung rejection has been more frequent and more problematic than cardiac rejection. Why might the lung be more prone to rejection? The lung is one of the largest

transplantable organs, with an extensive vasculature that is exposed to the entire cardiac output. The lung also has a vast intrinsic immune apparatus, which includes large populations of both antigen-presenting

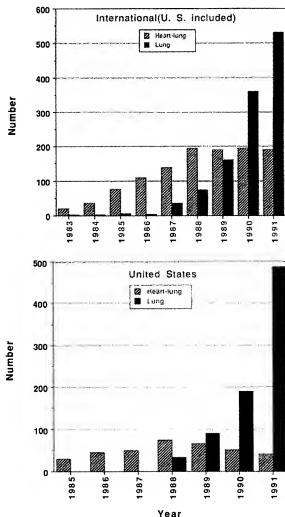


FIGURE 1. Top, International (United States included) lung and heart-lung transplant activity from 1983 through 1991. Data are from the St. Louis International Lung Transplant Registry (lung) and the International Society of Heart and Lung Transplantation (heart-lung).^{1,2} Bottom, United States lung and heart-lung transplant activity from 1985 through 1991. Data are from the United Network for Organ Sharing.³

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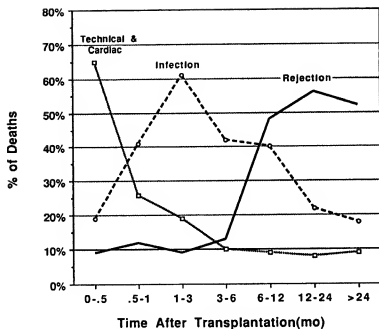


FIGURE 2. Causes of death after lung transplantation. Reproduced, with permission, from reference 1.

cells and effector cells, and the respiratory tract is in constant contact with extrinsic inhaled antigens, which may cause local inflammatory reactions with up-regulation of alloantigen expression on bronchial epithelium and activation of T lymphocytes.⁵

Lung transplantation has come of age,⁶ but many challenges remain. The immunobiology of the lung as an allograft has not been thoroughly characterized. Chronic rejection has been the major cause of late graft attrition and patient death, yet it is poorly understood and is difficult to treat. This article will review the management of lung transplant rejection from a clinical perspective. The mechanisms and classification of rejection will be presented, immunosuppressive strategies and drug regimens will be summarized, and diagnostic and therapeutic issues will be discussed.

MECHANISMS AND CLASSIFICATION OF REJECTION

A solid organ transplanted from a donor to a genetically nonidentical recipient is an allograft and provokes a specific immune response called rejection. The rejection reaction is an immunologic cascade leading to the activation, differentiation, and proliferation of effector T lymphocytes directed against donor cells. Transplants are rejected primarily because of differences between the donor and the recipient in cell-surface molecules that are encoded by genes in the major histocompatibility complex (MHC), commonly called the human leukocyte antigen (HLA) complex in humans. These polymorphic, cell-surface glycoproteins are divided into two major classes. Class I (HLA-A, B, C) molecules are constitutively expressed by most cells in most tissues. In contrast,

class II (HLA-DP, DQ, DR) molecules are constitutively expressed by relatively few cell types, such as B lymphocytes, macrophages, and monocytes; however, they can be induced on other cells, including T lymphocytes and endothelial cells, by interferon and other biologic response modifiers.^{7,9}

The physiologic function of MHC molecules is to present foreign antigens to T lymphocytes. T cells can respond only to antigens bound to the surface of antigen-presenting cells in association with class I or II MHC molecules. Helper T lymphocytes (CD4+) recognize antigens associated with class I MHC molecules, and cytotoxic T lymphocytes (CD8+) recognize those bound with class II molecules. The process by which donor-recipient MHC differences cause rejection is probably a variation of normal function in which donor MHC molecules that resemble recipient MHC-foreign antigen complexes become the target of cross-reactive recipient T lymphocytes.^{7,9}

Both donor class I and class II MHC molecules are alloantigens. Recipient cytolytic T lymphocytes (CD8+) recognize donor class I MHC molecules and, after differentiation under the influence of cytokines secreted from helper T lymphocytes, can directly lyse graft endothelial and parenchymal cells. Donor class II molecules on antigen-presenting cells such as macrophages stimulate recipient helper T lymphocytes (CD4+) to secrete interleukin-2 (IL-2) and other cytokines like gamma interferon. These cytokines act as costimulators, which induce MHC expression, enhance T lymphocyte responses, and amplify the cellular inflammatory reaction. In contrast to these forms of cell-mediated alloreactivity, considerably less is known about the pathogenesis of alloantibodies against

Table 1—Histologic Classification of Pulmonary Rejection*

Classification	Principal Histologic Feature
Acute rejection	Perivascular mononuclear infiltrates with or without airway inflammation
Active airway damage without fibrous scarring	Lymphocytic bronchitis or bronchiolitis without perivascular infiltrates
Chronic airway rejection	Bronchiolitis obliterans
Chronic vascular rejection	Fibrotic intimal thickening of arteries and veins
Vasculitis	Mononuclear infiltrate in walls of larger vessels

*Adapted from the working formulation of the Lung Rejection Study Group.¹¹

donor MHC molecules and the clinical significance of humoral rejection.

Graft rejection has traditionally been classified by its time of onset and its histopathologic pattern as hyperacute, acute, or chronic. Hyperacute rejection is caused by preexisting alloantibodies that bind to the vascular endothelium of the donor organ, activate complement, and cause rapid thrombosis of the graft vessels before the development of inflammation. Hyperacute rejection has been virtually eliminated by screening the serum of recipients for antibodies against a panel of cells representative of potential donors. When present, such antibodies may be the result of prior exposure to alloantigens through pregnancy, blood transfusion, or previous transplantation.

Hyperacute rejection after lung transplantation has not been documented but could occur. The constraint imposed by the relatively short acceptable ischemic time for the donor lungs precludes a direct, prospective crossmatch between the actual donor and the recipient. Thus, even though the recipient's serum had no panel-reactive antibodies, antibodies against donor alloantigens not represented in the screening panel could be present and could cause hyperacute rejection. However, only one case of presumed hyperacute rejection has been observed in 150 lung transplants performed at our center, and other cases have not been reported.

Acute rejection is a complex, integrated immune response stimulated by the recognition of alloantigens. Both specific cytotoxic T cell lysis and delayed-type hypersensitivity mechanisms probably effect graft injury, but the role of alloantibodies cannot be discounted.¹⁰ Histologically, acute rejection of the lung is characterized by perivascular mononuclear infiltrates with or without an accompanying lymphocytic bronchitis/bronchiolitis.¹¹ A classification scheme and a grading system for lung rejection are illustrated in Tables 1 and 2.¹¹

Chronic rejection has been the major cause of late graft attrition after solid organ transplantation, but it

is still an "undefined conundrum."¹² The common feature in all solid organs has been obliterative fibrosis of luminal structures in the allograft (eg, coronary arteries in the heart, bile ducts in the liver, and bronchioles in the lung). The pathogenesis of the lesion is uncertain, but hypotheses have included scarring caused by bouts of acute rejection, a fibrotic response to chronic ischemia related to vascular injury, and fibrosis generated by an ongoing subclinical immunologic insult.

In lung allografts, chronic rejection is a syndrome characterized histologically by bronchiolitis obliterans and physiologically by progressive airflow limitation. The lesion evolves from an inflammatory bronchiolitis to destruction and obliteration of the small airways. Mesenchymal cell growth factors may promote the fibroblastic response. Although infection, denervation, bronchial ischemia, diminished mucociliary clearance, and impaired lower respiratory tract defenses could all contribute to the development of bronchiolitis obliterans in the transplanted lung, airway-directed rejection is the most likely cause.^{13,14}

Lower respiratory tract infection has been postulated as a potential catalyst for obliterative bronchiolitis (OB).⁵ Lymphokines, such as interferon, released by the inflammatory reaction to the infection could augment the expression of MHC antigens and thereby trigger or magnify allograft rejection. Although this theory has not been verified, cytomegalovirus (CMV) pneumonitis has been postulated as a risk factor for the subsequent development of OB.¹⁵

IMMUNOSUPPRESSIVE STRATEGIES AND DRUG REGIMENS

Some rejection is almost inevitable after transplantation. Two strategies have been used to minimize or prevent rejection—making the graft less immunogenic and suppressing the recipient's immune response—but both of these approaches have practical limitations.

Table 2—Histologic Grading System for Acute Pulmonary Rejection*

Grade	Description
Normal (A0)	No significant abnormality
Minimal acute rejection (A1)	Infrequent perivascular infiltrates
Mild acute rejection (A2)	Frequent perivascular infiltrates around venules and arterioles
Moderate acute rejection (A3)	Dense perivascular infiltrates with extension into alveolar septa
Severe acute rejection (A4)	Diffuse perivascular, interstitial, and air-space infiltrates; alveolar pneumocyte damage; possibly parenchymal necrosis, infarction, or necrotizing vasculitis

*Adapted from the working formulation of the Lung Rejection Study Group.¹¹

Table 3—Immunosuppressive Drugs: Mechanisms of Action and Toxicities

Drug	Mechanisms of Action	Major Toxicities
Cyclosporine	Inhibits transcription of IL-2 gene; diminishes IL-2 production and release; blunts activation and proliferation of lymphocytes	Nephrotoxicity; hypertension; neurologic problems (tremors, paresthesias, headache, depression, confusion, seizures); hepatotoxicity; hypertrichosis; gingival hyperplasia
Azathioprine	Inhibits nucleic acid synthesis; blocks proliferation of lymphocytes	Leukopenia; pancreatitis; hepatitis and cholestatic jaundice
Corticosteroids	Decrease inflammatory reaction by lysing T lymphocytes and by blocking cytokine gene transcription and secretion from mononuclear phagocytes	Hyperglycemia; hypercholesterolemia; osteoporosis; cataracts; myopathy; peptic ulcer
ALG or ATG*	Opsonizes and depletes lymphocytes	Leukopenia; thrombocytopenia; fever; arthralgias; serum sickness
OKT3	Opsonizes or lyses lymphocytes with CD3 receptors, which are present on all mature T lymphocytes	Leukopenia; hypotension; pulmonary edema; aseptic meningitis; fever; chills; nausea, vomiting, and diarrhea; serum sickness

*ALG = antilymphocyte globulin; ATG = antithymocyte globulin; OKT3 = monoclonal antibody to the CD3 receptor on lymphocytes.

Hyperacute rejection has been virtually eliminated by ABO blood group compatibility matching between donor and recipient and by pretransplantation screening of recipients for panel-reactive antibodies. Theoretically, cellular rejection can be reduced by minimizing differences between donor and recipient at the MHC (HLA) loci. In kidney transplantation, tissue typing and donor-recipient HLA matching have been beneficial and are standard practice. In other solid organs, including the lung, the importance of HLA matching has not been extensively studied, and the time limit on organ preservation has precluded routine HLA matching between the donor and the recipient. In an analysis of the early heart-lung transplant experience at Stanford University Medical Center, recipients with zero to one mismatch at HLA locus A had a tendency toward less OB, less severe OB, and fewer deaths due to OB than recipients with two mismatches at this locus,¹⁶ but no other studies have investigated this link.

The mainstay of solid organ transplantation has been drugs that suppress the recipient's immune response. The commonly used agents are summarized

in Table 3. Although these drugs effectively modulate reactivity to the allograft, they are nonspecific immune suppressants and predispose recipients to both opportunistic and nonopportunistic infections. Furthermore, immunologic monitoring of the effects of these drugs is not yet clinically applicable, and therapy is regulated by evidence of drug toxicity and rejection.

The protocols for immunosuppressive therapy can be divided into three general categories: induction, maintenance, and treatment of rejection. In lung transplantation, these regimens have been adopted or modified from the experience in other solid organ transplants. The protocols in use in our program on July 1, 1992 are illustrated in Table 4. A three-drug regimen of cyclosporine, azathioprine, and a corticosteroid is now being used for both induction and maintenance, but the use of corticosteroids during the first 7-10 days after transplantation has evolved with experience. During the induction phase antilymphocyte globulin has been added for more intensive immunosuppression.

Since its introduction in 1981, cyclosporine has become the cornerstone of most immunosuppressive

Table 4—Immunosuppressive Drug Regimens*

Phase	Drug	Regimen
Induction	Cyclosporine	2-4 mg/h intravenously initially, then adjust to obtain blood level in high therapeutic to slightly supratherapeutic range; gradually change to oral therapy when feasible (bioequivalent oral doses are about 3-4 times larger than IV dose)
	Azathioprine	2 mg/kg/d intravenously and then orally
	Corticosteroids	Methylprednisolone, 1 g, intravenously intraoperatively before reperfusion; methylprednisolone, 1 mg/kg/d, intravenously for the first 3 days, then IV methylprednisolone or oral prednisone, 0.5 mg/kg/d
Maintenance	Antilymphocyte globulin	10-15 mg/kg/d intravenously for the first 5-7 days only
	Cyclosporine	Titrate dose according to trough blood level; target levels in the mid-to-upper therapeutic range, but modified appropriately for nephrotoxicity
	Azathioprine	2 mg/kg/d, increase dose to 2.5-3 mg/kg/d for recurrent acute or chronic rejection; decrease dose for leukocyte count <4,500/mm ³
	Prednisone	0.5 mg/kg/d for the first 3 months; taper to 15 mg/d by 6 months, then to 15 mg on alternate days by 12 months, and maintain there indefinitely

*Regimens in use in the Washington University/Barnes Hospital Lung Transplant Program on July 1, 1992. IV = intravenous.

drug regimens.¹⁷ The clinical pharmacology is complex, but cyclosporine is metabolized by the hepatic cytochrome P-450 system, and its clearance may be altered by other drugs that interact with this system. Commonly used drugs that inhibit cytochrome P-450 and increase cyclosporine levels include ketoconazole, erythromycin, oral contraceptives, methylprednisolone, and the calcium channel antagonists diltiazem and verapamil, but not nifedipine. Likewise, drugs that induce P-450 enzymes tend to lower cyclosporine levels; potential effects may be produced by rifampin, phenobarbital, phenytoin, carbamazepine, and valproic acid.

Because of the variability in cyclosporine pharmacokinetics, monitoring the drug level is crucial to optimum therapy. Trough levels should be measured using an assay directed primarily at the active parent compound. In the early posttransplantation period, cyclosporine levels are usually kept in the high therapeutic to somewhat supratherapeutic range unless the serum creatinine concentration rises unacceptably. Subsequently, target levels are in the mid-to-upper therapeutic range in the absence of nephrotoxicity.

Azathioprine has been a standard drug in transplantation since the 1960s. Its relevance in lung transplantation was confirmed by the Stanford experience with their initial group of heart-lung transplant recipients. These recipients were managed with a two-drug regimen of cyclosporine and prednisone, and the incidence of OB was over 50 percent.^{14,18,19} Augmented immunosuppression with azathioprine blunted the rate of progression of OB in afflicted patients, and the incidence of OB has been reduced to about 20 percent in the subsequent group of recipients treated with a three-drug protocol including azathioprine. Although other changes in management, such as more intensive surveillance, may have contributed to the decline in OB, the role of azathioprine in lung transplantation was firmly established by this experience.

While the use of cyclosporine and azathioprine has been relatively uniform, corticosteroid usage for induction and early maintenance therapy has changed considerably over the past 10 years. Because of the historical problem with airway dehiscence in the series of human lung transplants between 1963 and 1973 and the detrimental effect on the strength of the bronchial anastomosis in a canine experimental model,^{20,21} pretransplantation and early posttransplantation corticosteroid therapy were initially avoided except for the treatment of acute rejection. Recent results, however, have allayed this concern,²² and although dosages differ, corticosteroids are now part of the induction regimen at most centers. Prednisone is usually included in the maintenance drug regimen, at least initially, but both the dosage and the taper schedule vary among programs. Low-dose prednisone is contin-

ued indefinitely in our recipients, but the transplant group at Papworth Hospital has reported good long-term results in heart-lung transplant recipients without maintenance prednisone.²³

Antilymphocyte antibodies have been used for induction therapy and for the treatment of refractory acute or chronic rejection, but their role in lung transplantation has not been clearly delineated. The available preparations include the polyclonal antisera, such as antilymphocyte globulin (ALG) and antithymocyte globulin (ATG), and the monoclonal antibody to the CD3 receptor on lymphocytes, OKT3. Antilymphocyte globulin has been a standard component of our induction regimen, but satisfactory results with OKT3 have also been reported.²⁴ However, the risk of postoperative CMV infection may be significantly increased by cytolytic therapy with either ALG or OKT3.²⁵ Both the efficacy and the complications of these agents need further study.

Among the investigational drugs, FK 506, a fungal macrolide with potent immunosuppressive effects, has received the greatest attention. Clinical trials have focused on liver transplantation, but a small number of other solid organ recipients have also been treated.²⁶ Experience in heart-lung and lung transplant recipients is very limited and inconclusive. At the present time, FK 506 is not readily available for clinical use after lung transplantation.

DIAGNOSIS AND TREATMENT

Pulmonary rejection has been common after both heart-lung and isolated lung transplantation, and the rejection pattern has been similar after each of these procedures.²⁷ Most recipients have at least one episode of acute rejection, and about 25 percent of recent long-term survivors have developed chronic rejection. Acute rejection is usually readily reversible with treatment and is rarely fatal. In contrast, chronic rejection often responds poorly to therapy and causes considerable late morbidity and mortality.

Appropriate management of rejection is dependent on timely, accurate diagnosis. Theoretically, the diagnosis of rejection could be approached clinically, histologically, or immunologically. Unfortunately, sensitive and specific immunologic tests for monitoring the adequacy of immunosuppressive therapy or for detecting allograft rejection are not currently available. Thus, clinical and histologic criteria are currently the diagnostic standards.

Acute Rejection

Acute rejection has been observed as early as 3 days and as late as several years after transplantation. Using clinical criteria, nearly all of our lung transplant recipients have had at least one instance of acute rejection during the first 3 weeks; using a decision-to-

treat analysis, the average number of episodes per patient during this time interval has been approximately 2.5. Using histologic criteria, 60 percent of heart-lung recipients at Papworth Hospital had a biopsy-proven episode during the first postoperative month, and 60 percent of all cases of acute rejection occurred during the first 3 months after transplantation.²³ Although the likelihood of acute rejection appears to decrease over time, the risk is always present, and late episodes are not unusual.

Acute rejection has many manifestations; early (<1 month after transplantation) and late episodes may present somewhat differently. Clinical features include cough, dyspnea, fever, adventitious lung sounds (rales, wheezes), radiographic infiltrates, hypoxemia, and deterioration in pulmonary function test (PFT) results. Some typical diagnostic guidelines are summarized in Table 5. Obviously, the clinical profile of acute pulmonary rejection is nonspecific. Within the first few weeks after transplantation, a comparable clinical picture can be caused by postoperative purulent bronchitis with retained secretions, pneumonia, pulmonary edema, or posttransplantation acute lung injury (reimplantation response). Beyond the first month, infection is the main alternative in the differential diagnosis. Cytomegalovirus pneumonia, the most common opportunistic infection, generally cannot be distinguished from rejection without a biopsy.

The chest radiograph is usually abnormal during acute rejection in the first month after transplantation,

Table 5—Clinical Criteria for the Diagnosis of Rejection

Temperature: Rise >0.5°C above stable baseline
Oxygenation: Fall >10 mm Hg below stable baseline
Radiograph: New or changing infiltrates
Spirometry: Fall in FEV ₁ >10% below stable baseline
Infection excluded
Response to treatment with methylprednisolone

but is seldom abnormal thereafter. In a series of heart-lung recipients with biopsy-confirmed rejection, the chest radiograph was abnormal in 74 percent of the episodes during the first month, but was normal in 77 percent of those episodes that occurred later than 1 month after transplantation.²⁴ The most frequent radiographic pattern has been the combination of parenchymal shadows (perihilar and lower lung zone infiltrates and/or septal lines) and pleural effusions,^{25,26} but these radiographic changes are not unique (Fig 3, 4). Cytomegalovirus pneumonitis can produce similar abnormalities, including interstitial infiltrates, septal lines, and small effusions.^{26,27}

Although pulmonary function is abruptly improved by transplantation, the recovery reflected by PFT values continues for 1 to 3 months after transplantation as the perioperative changes in the lung and thoracic cage resolve. The final PFT pattern is determined by the underlying disease and the type of operation. The PFT values revert to normal after heart-lung and bilateral transplantation, but mild-to-moderate abnormalities that reflect the pathophysiology of the re-

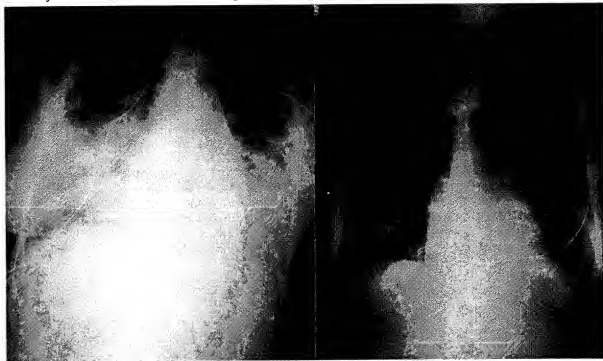


FIGURE 3. Left, Chest radiograph of a bilateral lung transplant recipient with acute rejection 5 days after transplantation. Right, Chest radiograph of the same recipient 8 h after treatment with methylprednisolone.

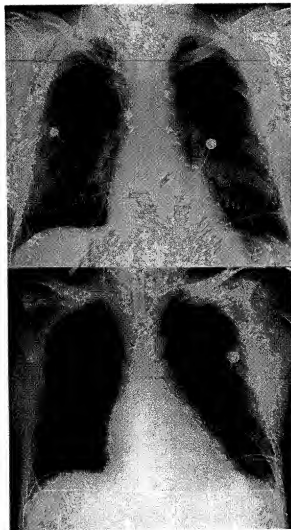


FIGURE 4. Top, Chest radiograph of a bilateral lung transplant recipient with a pattern mimicking acute rejection 5 days after transplantation. Bronchoscopy revealed retained mucopurulent secretions throughout the bronchial tree. Bottom, Chest radiograph of the same recipient 6 h after clearance of the secretions by bronchoscopy.

maintaining native lung persist after single lung transplantation. Nonetheless, once perioperative changes have resolved and lung function has stabilized, the coefficient of variation for most PFT parameters is quite small, usually no more than 5 percent.³⁰ Thus, a decline of 10 percent or more in forced vital capacity (FVC) and/or FEV₁ is a significant change and may signal a pulmonary complication.³¹

Oxygenation and PFT values deteriorate during both acute rejection and infection. Although sensitive for detecting either infection or rejection, changes in PFT values have not discriminated between these complications.³⁰ The FVC and FEV₁ fell proportionately with either infection or rejection in one study of heart-lung recipients,³⁰ but a pattern of small airway

obstruction was more suggestive of rejection in another investigation.³² In our own series of lung transplant recipients,³³ the development of expiratory airflow limitation has been more suggestive of rejection than CMV pneumonia.

Because clinical criteria alone cannot reliably differentiate between rejection and infection, a more definitive approach must be used. In other solid organ grafts, biopsy has been the conventional method for diagnosing rejection. Although originally deemed inadequate in a canine model,³⁴ transbronchial biopsy (TBB) has emerged as the procedure of choice for diagnosing rejection and infection in lung transplant recipients.³⁵⁻³⁹ A histologic classification scheme and a grading system for acute rejection have been formulated (Tables 1, 2).¹¹ Acute rejection is identified by perivascular, mononuclear infiltrates, which may be accompanied by a lymphocytic bronchitis/bronchiolitis; in more severe rejection, the inflammation also extends into the surrounding parenchyma. Although perivascular inflammation has been considered almost pathognomonic of rejection in lung transplant recipients, it has been described in CMV pneumonia and *Pneumocystis carinii* pneumonia.⁴⁰ Thus, rejection must be interpreted cautiously if there is evidence of either CMV or *Pneumocystis* infection.

When performed for a clinical indication such as differentiating suspected rejection from infection, TBB has had a positivity rate of 69 to 83 percent.^{32,36,39} The sensitivity for diagnosing rejection has ranged from 72 to 94 percent; the specificity, from 90 to 100 percent.^{35,38,39} The diagnostic yield has been good with an average of 4 to 10 biopsies per procedure,^{38,39} but 18 biopsies have been suggested to reach the 95 percent confidence limit for the detection of rejection.³⁸

The complication rate of TBB has been acceptably low, overall rates varying from 9 to 18 percent.^{30,39} Pneumothorax, the most common serious complication of TBB, has occurred infrequently, probably because of postoperative pleural adhesions. Excessive bleeding has been the most frequent complication, but the incidence has been lower than reported in other immunocompromised patients.

Bronchoalveolar lavage (BAL) is a useful adjunct to TBB, but its clinical utility is confined to the diagnosis of infection after lung transplantation. Simple differential counts of the cellular constituents of BAL fluid have not segregated rejection from infection,⁴¹ however, BAL lymphocytes harvested during episodes of acute rejection have manifested increased donor-specific alloreactivity in a primed lymphocyte test.⁴² Although the primed lymphocyte assay has a high sensitivity, it lacks the specificity to exclude infection, and it requires a preserved supply of donor cells and a 72-h incubation period.

Standard treatment for acute pulmonary rejection is high-dose corticosteroids, but the maintenance immunosuppressive regimen should also be reviewed and optimized. Methylprednisolone, 500 to 1,000 mg/d for 3 days, is the conventional regimen, and this is sufficient to reverse most episodes. If the maintenance prednisone therapy has been decreased or stopped, escalating the dose to approximately 1 mg/kg/d and tapering it down again over 2 to 3 weeks may be beneficial in more severe early episodes or in later episodes. Although steroid-resistant acute rejection is unusual, refractory acute rejection has been successfully treated with OKT3.⁴³

After successful treatment, the radiographic, physiologic, and histologic abnormalities improve. However, the time course for complete resolution of cellular infiltrates has not been fully determined. Small, persistent perivascular infiltrates have been described on follow-up biopsies 3 weeks after treatment in two thirds of recipients.⁴⁴ When present, peribronchial infiltrates have shown less response during this interval.⁴⁴ Should patients with residual perivascular or peribronchial infiltrates be retreated? At the present time, this question cannot be answered decisively, but probably not if the biopsy specimen shows improvement and the physiologic status has returned toward baseline.

Chronic Rejection

Chronic rejection after lung transplantation has become virtually synonymous with OB. Posttransplant OB is a clinicopathologic syndrome characterized physiologically by airflow limitation and histologically by bronchiolitis obliterans and variable vascular sclerosis. It is the most problematic late complication of lung transplantation and the leading cause of late mortality (Fig 2). With contemporary maintenance immunosuppressive drug regimens and surveillance protocols, the prevalence is still 20 to 40 percent in long-term survivors of heart-lung and isolated lung transplantation, and the case fatality rate has remained high.^{16,45-47} The causes of death have been progressive respiratory failure and infection, often complicating intensive immunosuppressive therapy for OB.

Two potential risk factors for the development of OB—antecedent acute rejection and CMV infection—have been proposed. Pulmonary infection has frequently been a forerunner of OB, and CMV infection has been implicated as a risk factor for OB.^{15,48} However, OB has occurred without preceding CMV infection, and the association between CMV infection and OB has not always been confirmed.^{33,49} Chronic rejection has also been linked to acute rejection.^{45,49,50} Recipients with more severe, more frequent, and more persistent acute rejection episodes have had a significantly higher relative risk for chronic rejection.

Obliterative bronchiolitis has been diagnosed as early as 2 months and as late as several years after transplantation, but in most series the mean time to development has been approximately 8 to 12 months.^{15,19,46,48} The presentation is variable, but two patterns have been discerned. The most common presentation mimics an upper respiratory tract infection or bronchitis, and, indeed, OB may be triggered by or evolve after a viral respiratory infection.⁵ Such illnesses always raise the specter of rejection and must be scrutinized and monitored carefully. Less often, the early stage of OB evolves more insidiously and is manifested primarily by progressive decrements in FEV₁ on surveillance PFTs.

The diagnosis of OB is based on clinical, physiologic, and pathologic aspects. The evolution of new or worsening airflow limitation is strongly suggestive of rejection; after a stable baseline has been established, a decline in FEV₁ of more than 10 percent is usually significant. When OB is suspected, the diagnosis should be confirmed histologically if possible, but the sensitivity of TBB for detecting OB has been highly variable, ranging from 5 to 100 percent.^{49,51,52} Moreover, the specificity of the pathologic findings alone has been low; comparable histologic abnormalities, presumably related to previous episodes of infection or rejection, have been present in the biopsy specimens of recipients without the clinical features of OB.^{45,49} However, if both the clinical picture and the PFT pattern are compatible and other causes have been eliminated, OB is almost always the diagnosis. Open lung biopsy is not necessary unless this conclusion is in doubt.

Prevention of OB is the goal, of course, but while the incidence has decreased, current strategies have not abolished the problem. Protection is based on an optimal maintenance immunosuppressive drug regimen; however, an ideal regimen has not been determined. Furthermore, maintenance therapy is often compromised by drug toxicity, and extrinsic factors may trigger rejection in spite of otherwise adequate immunosuppression. Because acute rejection and infection have been incriminated as precursors of chronic rejection, their prompt diagnosis and treatment could also influence the evolution of chronic rejection.

Chronic rejection has been treated by augmenting the immunosuppressive drug regimen with high-dose corticosteroids, azathioprine, and the antilymphocyte agents ALG, ATG, and OKT3. Because of its impact on OB,¹⁸ azathioprine has now been incorporated into most maintenance drug protocols. Typical corticosteroid therapy has been methylprednisolone, approximately 1 gm/d for 3 days, followed by prednisone, approximately 1 mg/kg/d initially with a taper to the pretreatment dosage over 2 to 3 weeks. Both ALG and

ATG have generally been given for 10 to 14 days; the standard dosage for the most widely available preparations, Minnesota ALG and ATGAM (Upjohn), is 10 to 20 mg/kg/d, but other institutionally formulated antisera may have a different dosage. The recommended treatment regimen with OKT3 is 5 mg/d for 10 to 14 days.

Beneficial results—either stabilization or improvement in PFT values—have been achieved with corticosteroids and the antilymphocyte preparations, but the relapse rate has been high after both forms of treatment.⁴⁷ Overall response rates of 65 percent for corticosteroids and 81 percent for antilymphocyte agents have recently been reported, but 69 percent of corticosteroid-treated and 55 percent of antilymphocyte agent-treated episodes eventually relapsed.⁴⁷

During and after intensive immunosuppression for chronic rejection, the risk of infection is substantially increased. A course of therapy should not be initiated until any active infection has been controlled or eradicated. Prophylaxis for CMV infection in recipients at risk and treatment of noninvasive *Aspergillus* infection in recipients colonized by this organism should be considered, especially during treatment with the antilymphocyte drugs.

SURVEILLANCE

Because of the ongoing risk of rejection, routine surveillance schemes have been implemented by many centers. Most plans have involved early reporting of symptoms, daily home spirometry, regular PFTs, and periodic bronchoscopy with TBB, but the schedule for PFTs and TBB has not been uniform among centers. In our program PFTs have been done weekly for the first 3 months after transplantation and then monthly for the remainder of the first year. Thereafter, PFTs have been repeated every 2 to 3 months. Surveillance TBB has been done initially about 2 to 3 weeks and again 8 to 12 weeks after transplantation, but more frequent biopsies may be appropriate during this period. Subsequently, TBB has been repeated at 6 months, 1 year, and then annually after transplantation.

Small, affordable spirometers have made monitoring graft function at home practical. These spirometers can reproducibly measure FVC and FEV₁, and some models can also record other expiratory parameters. Downward trends in FVC or FEV₁ of 10 percent or more that persist for several days may signal a pulmonary complication, but neither the magnitude nor the pattern of change can reliably separate infection and rejection.³¹

The best use of TBB for graft surveillance has not been defined, but the available data have raised some important questions and concerns. The positivity rate of surveillance TBB procedures has ranged from 15 to

57 percent, and both acute rejection and infection have been discovered in asymptomatic, clinically and physiologically stable recipients.^{32,39} Low-grade (minimal or mild) rejection has been recorded in 33 to 40 percent of procedures,^{30,33} and histologic evidence of occult CMV infection has been found in up to 17 percent of cases.³⁹ Although these clinically silent findings are worrisome, their ultimate impact is uncertain. The outcome of low-grade untreated rejection in a small group of clinically well heart-lung recipients was not unfavorable,³⁰ and the natural history of occult CMV infection/pneumonia is unknown. Because acute rejection and CMV pneumonia may be related to the development of chronic rejection, our policy has been to treat histologically documented rejection (\geq grade A2) or CMV infection (cells with characteristic inclusions, with or without associated pneumonitis) found in surveillance biopsy specimens.

SUMMARY

Using current immunosuppressive protocols, rejection is common after lung transplantation. Most recipients have at least one episode of acute rejection, and approximately 25 percent of recent long-term survivors have developed chronic rejection. Acute rejection has usually been reversible with treatment, but chronic rejection has responded poorly, relapsed frequently, and been one of the leading causes of late morbidity and mortality.

Appropriate management of rejection is predicated on timely, accurate diagnosis. Clinical criteria for the diagnosis of acute rejection are useful but nonspecific, and TBB has emerged as the procedure of choice for diagnosing acute rejection and infection. Chronic rejection is manifested by OB and is characterized physiologically by the development of airflow obstruction. Although histologic confirmation is preferable, the sensitivity of TBB for the detection of OB has been inconsistent, and the specificity has been low.

Lung transplantation has indeed come of age, but understanding the immunopathogenesis and improving the clinical management of rejection remain major challenges for the next decade.

REFERENCES

- 1 Kriett JM, Kaye ME. The registry of the International Society of Heart and Lung Transplantation: eighth official report—1991. *J Heart Lung Transplant* 1991; 10:491-98
- 2 Cooper JD. St. Louis International Lung Transplant Registry Interim Report, August 1992
- 3 United Network for Organ Sharing. UNOS releases 1991 transplant statistics. *UNOS Update* 1992; 8(5):1-2
- 4 Paradis I, Rabinowich H, Zeveloff A, Yousem S, Noyes B, Hoffman R, et al. Life in the allogeneic environment after lung transplantation. *Lung* 1990; suppl:1173-81
- 5 Burke CM, Glanville AR, Theodore J, Robin ED. Lung immunogenicity, rejection, and obliterative bronchiolitis. *Chest* 1987; 92:547-49

- 6 Theodore J, Lewiston N. Lung transplantation comes of age. *N Engl J Med* 1990; 322:772-74
- 7 Bach F, Sachs DH. Transplantation immunology. *N Engl J Med* 1987; 317:489-92
- 8 Krensky AM, Weiss A, Crahtree C, Davis MM, Parham P. T-lymphocyte-antigen interactions in transplant rejection. *N Engl J Med* 1990; 322:510-17
- 9 Abbas AK, Lichtman AH, Pober JS. Immunity to tissue transplants. In: Cellular and molecular immunology. Philadelphia: WB Saunders, 1991:317-34
- 10 Bradley JA, Bolton EM. The T-cell requirements for allograft rejection. *Transplant Rev* 1992; 6:115-29
- 11 Yousem SA, Berry CJ, Brunt EM, Chamberlain D, Hriban RH, Sibley RK, et al. A working formulation for the standardization of nomenclature in the diagnosis of heart and lung rejection: Lung Rejection Study Group. *J Heart Transplant* 1990; 9:593-601
- 12 Tilney NL, Whitley WD, Diamond JR, Kupiec-Weglinski JW, Adams DH. Chronic rejection: an undefined conundrum. *Transplantation* 1991; 52:389-98
- 13 Griffith BP, Paradis IL, Zeevi A, Rabinowich H, Yousem SA, Duquesnoy RJ, et al. Immunologically mediated disease of the airways after pulmonary transplantation. *Ann Surg* 1988; 208:371-78
- 14 Theodore J, Starnes VA, Lewiston NJ. Obliterative bronchiolitis. *Clin Chest Med* 1990; 11:309-21
- 15 Keenan RJ, Lega ME, Dummer JS, Paradis IL, Dauber JH, Rabinowich H, et al. Cytomegalovirus serologic status and postoperative infection correlated with risk of developing chronic rejection after pulmonary transplantation. *Transplantation* 1991; 51:433-38
- 16 Harjula ALJ, Baldwin JC, Claville AR, Tazelaar H, Oyer PE, Stinson EB, et al. Human leukocyte antigen compatibility in heart-lung transplantation. *J Heart Transplant* 1987; 6:162-66
- 17 Kahan BD. Cyclosporine. *N Engl J Med* 1989; 321:1725-38
- 18 Claville AR, Baldwin JC, Burke CM, Theodore J, Robin ED. Obliterative bronchiolitis after heart-lung transplantation: apparent arrest by augmented immunosuppression. *Ann Intern Med* 1987; 107:300-04
- 19 McCarthy PM, Starnes VA, Theodore J, Stinson EB, Oyer PE, Shumway NE. Improved survival after heart-lung transplantation. *J Thorac Cardiovasc Surg* 1990; 99:54-60
- 20 Wildevuur CRH, Benfield JR. A review of 23 human lung transplantations by 20 surgeons. *Ann Thorac Surg* 1970; 9:489-515
- 21 Lima O, Cooper JD, Peters WJ, Ayabe H, Townsend E, Luk SC, et al. Effects of methylprednisolone and azathioprine on bronchial healing following lung autotransplantation. *J Thorac Cardiovasc Surg* 1981; 82:211-15
- 22 Calhoun JH, Grover FL, Gibbons WJ, Bryan CL, Levine SM, Bailey SR, et al. Single lung transplantation: alternative indications and technique. *J Thorac Cardiovasc Surg* 1991; 101:816-25
- 23 Hutter JA, Despins P, Higenbottam T, Stewart S, Wallwork J. Heart-lung transplantation: better use of resources. *Am J Med* 1988; 85:4-11
- 24 Waters P. Experience with OKT3 and human lung transplantation [abstract]. *Chest* 1990; 98(Suppl):75
- 25 Calhoun JH, Nichols L, Davis R, Bryant CL, Levine SM, Zamora CA, et al. Single lung transplantation: factors in post-operative cytomegalovirus infection. *J Thorac Cardiovasc Surg* 1992; 103:21-6
- 26 Todo S, Fung JJ, Starzl TE, Trakis A, Demetris AJ, Korman R, et al. Liver, kidney, and thoracic organ transplantation under FK 506. *Ann Surg* 1990; 212:285-307
- 27 Keenan RJ, Bruzzone E, Paradis IL, Yousem SA, Dauber JH, Stuart RS, et al. Similarity of pulmonary rejection patterns among heart-lung and double-lung transplant recipients. *Transplantation* 1991; 51:176-80
- 28 Millet B, Higenbottam TW, Flower CDR, Stewart S, Wallwork J. The radiographic appearances of infection and acute rejection of the lung after heart-lung transplantation. *Am Rev Respir Dis* 1989; 140:62-7
- 29 Bergin CJ, Castellino RA, Blank N, Berry CJ, Sibley RK, Starnes VA. Acute lung rejection after heart-lung transplantation: correlation of findings on chest radiographs with lung biopsy results. *AJR* 1990; 155:23-7
- 30 Otulana BA, Higenbottam T, Scott J, Clelland C, Igboaka G, Wallwork J. Lung function associated with histologically diagnosed acute lung rejection and pulmonary infection in heart-lung transplant patients. *Am Rev Respir Dis* 1990; 142:320-32
- 31 Otulana BA, Higenbottam T, Ferrar L, Scott J, Igboaka G, Wallwork J. The use of home spirometry in detecting acute lung rejection and infection following heart-lung transplantation. *Chest* 1990; 97:353-57
- 32 Starnes VA, Theodore J, Oyer PE, Billingham ME, Sibley RK, Berry C, et al. Evaluation of heart-lung transplant recipients with prospective, serial transbronchial biopsies and pulmonary function studies. *J Thorac Cardiovasc Surg* 1989; 98:683-90
- 33 Ettinger NA, Bailey TC, Trulock EP, Storch GA, Anderson D, Raah S, et al. Cytomegalovirus pneumonitis: impact following isolated lung transplantation. *Am Rev Respir Dis* (in press)
- 34 Koerner SK, Hagstrom JWC, Veith FJ. Transbronchial biopsy for the diagnosis of lung transplant rejection: comparison with needle and open lung biopsy techniques in canine lung allografts. *Am Rev Respir Dis* 1976; 114:575-79
- 35 Higenbottam T, Stewart S, Penketh A, Wallwork J. Transbronchial lung biopsy for the diagnosis of rejection in heart-lung transplant patients. *Transplantation* 1988; 46:532-39
- 36 Starnes VA, Theodore J, Oyer PE, Stinson EB, Moreno-Cabral CE, Sibley R, et al. Pulmonary infiltrates after heart-lung transplantation: evaluation by serial transbronchial lung biopsies. *J Thorac Cardiovasc Surg* 1989; 98:945-50
- 37 Marchevsky A, Hartman G, Walts A, Ross D, Koerner S, Waters P. Lung transplantation: the pathologic diagnosis of pulmonary complications. *Mod Pathol* 1991; 4:133-38
- 38 Scott JP, Fradet G, Smyth RL, Mullins P, Pratt A, Clelland CA, et al. Prospective study of transbronchial biopsies in the management of heart-lung and single lung transplant patients. *J Heart Lung Transplant* 1991; 10:626-37
- 39 Trulock EP, Ettinger NA, Brunt EM, Pasque MK, Kaiser LR, Cooper JD. The role of transbronchial lung biopsy in the treatment of lung transplant recipients: an analysis of 200 consecutive procedures. *Chest* 1992; 102:1049-54
- 40 Tazelaar HD. Perivascular inflammation in pulmonary infections: implications for the diagnosis of lung rejection. *J Heart Lung Transplant* 1991; 10:437-41
- 41 Ettinger NA, Tsugi H, Muthupackal J, Trulock EP. The use of bronchoalveolar lavage (BAL) to distinguish infection from rejection in single and double lung transplantation [abstract]. *Am Rev Respir Dis* 1991; 143A, p2:A600
- 42 Rabinowich H, Zeevi A, Paradis IL, Yousem SA, Dauber JH, Korman R, et al. Proliferative responses of bronchoalveolar lavage lymphocytes from heart-lung transplant patients. *Transplantation* 1990; 49:115-21
- 43 Shennib H, Mercado M, Nguyen D, Ernst E, Lebel F, O'Donovan M, et al. Successful treatment of steroid-resistant double-lung allograft rejection with Orthoclone OKT3. *Am Rev Respir Dis* 1991; 144:224-26
- 44 Clelland CA, Higenbottam TW, Stewart S, Scott JP, Wallwork J. The histological changes in transbronchial biopsy after treatment of acute lung rejection in heart-lung transplant recipients. *J Pathol* 1990; 161:105-12
- 45 Scott JP, Higenbottam TW, Sharples L, Clelland CA, Smyth

- RL, Stewart S, et al. Risk factors for obliterative bronchiolitis in heart-lung transplant recipients. *Transplantation* 1991; 51:813-17
- 46 Anzueto A, Levine SM, Bryan CL, Peters JI, Zamora CA, Calhoun JH, et al. Obliterative bronchiolitis in single lung transplant recipients [abstract]. *Am Rev Respir Dis* 1992; 145(4; pt 2):A700
- 47 Paradis IL, Duncan SR, Dauber JH, Costantino JP, Similo S, Yousem SA, et al. Effect of augmented immunosuppression on human chronic lung allograft rejection [abstract]. *Am Rev Respir Dis* 1992; 145(4; pt 2):A705
- 48 Cerrina J, Le Roy LF, Parquin F, Rain B, Simmoneau G, Chapelier A, et al. Risk factors for obliterative bronchiolitis (OB) after heart-lung (HLT) and double lung transplantation (DLT) [abstract]. *Am Rev Respir Dis* 1992; 145(4; pt 2):A700
- 49 Scott JE, Higenbottam TW, Clelland CA, Stewart S, Smyth RL, McGoldrick JP, et al. Natural history of chronic rejection in heart-lung transplant recipients. *J Heart Transplant* 1990; 9:510-15
- 50 Yousem SA, Dauber JA, Keenan R, Paradis IL, Zeevi A, Griffith BP. Does histologic acute rejection in lung allografts predict the development of bronchiolitis obliterans? *Transplantation* 1991; 52:306-09
- 51 Yousem SA, Paradis IL, Dauber JH, Griffith BP. Efficacy of transbronchial lung biopsy in the diagnosis of bronchiolitis obliterans in heart-lung transplant recipients. *Transplantation* 1989; 47:893-95
- 52 Kramer MR, Sibley R, Stoeck C, Starnes VA, Theodore J. Transbronchial lung biopsies have low yield in the detection of post heart-lung and lung transplantation obliterative bronchiolitis [abstract]. *Am Rev Respir Dis* 1992; 145(4; pt 2):A699
- 53 Clelland C, Higenbottam T, Otulana B, Stewart S, Igboaka G, Scott J, et al. Histologic prognostic indicators for the lung allografts of heart-lung transplants. *J Heart Transplant* 1990; 9:177-86